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Product Information: **ATTO 495/665**



ATTO 495/665 is a new fluorescent label featuring an extraordinary large **Stokes-Shift** of **170 nm**. Thus its emission is spectrally almost completely separated from its absorption. **ATTO 495/665** is very hydrophilic and highly soluble. Emitting at such a long wavelength the dye exhibits a high fluorescence quantum yield, which is most important only slightly reduced after conjugation to a biomolecule e.g. proteins. This is even the case at a high „Degree of Labeling (DOL)“.

ATTO 495/665 is an anionic dye. After conjugation to a substrate the dye carries a net electrical charge of -1. For details of coupling see our recommended labeling procedure at www.atto-tec.com - Support - Downloads - [General Procedures](#).

Optical data of the carboxy derivative (in water):

$$\lambda_{\text{abs}} = 495 \text{ nm}$$

$$\epsilon_{\text{max}} = 4.0 \times 10^4 \text{ M}^{-1} \text{ cm}^{-1}$$

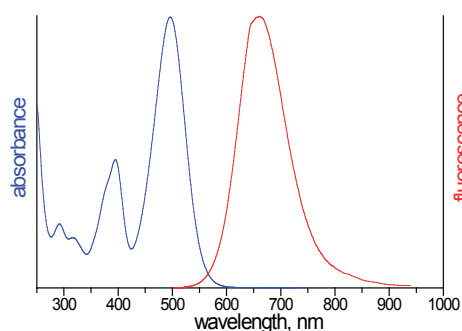
$$\lambda_{\text{fl}} = 665 \text{ nm}$$

$$\eta_{\text{fl}} = 30 \%$$

$$\tau_{\text{fl}} = \text{x.x ns}$$

$$\text{CF}_{260} = 0.37$$

$$\text{CF}_{280} = 0.18$$



Spectra available in digitized form (excel file) on <http://www.atto-tec.com>

Modification	MW, g/mol	M ⁺ , g/mol	Order Code	
			Unit (1 mg)	Unit (5 mg)
with free COOH	696	674	AD 495/665-21	AD 495/665-25
NHS-ester	794	772	AD 495/665-31	AD 495/665-35
maleimide	818	796	AD 495/665-41	AD 495/665-45
phalloidin	xxx	796	AD 495/665-81*	AD 495/665-82**
azide	xxx	796	AD 495/665-101	AD 495/665-105

* 10 nmol **20 nmol

General Information

Storage: The product is shipped solvent-free at ambient temperature. Upon receipt store at -20 °C. To avoid moisture condensation onto the product, vial must be equilibrated to room temperature before opening. When stored properly, protected from moisture and light, ATTO-TEC products are stable for at least three years.

Risk and safety: A material safety data sheet (MSDS) of each derivative can be downloaded from our website at www.atto-tec.com.

Solutions: The product is soluble in polar solvents, e.g. dimethylformamide (DMF), dimethylsulfoxide (DMSO), or water. However, due to their inherent reactivity, NHS-esters and maleimides must be well protected from OH-containing solvents like ethanol and, in particular, water. Prepare labeling solutions of NHS-esters and maleimides immediately before use by dissolving the vial content in anhydrous and amine-free DMF or DMSO. Depending on the quality of the solvent used, such solutions may be of limited stability.

Dye with **free COOH** may be used for any kind of spectroscopy. Due to the high extinction coefficient and its high quantum yield of fluorescence this product is suitable for high-sensitivity detection including single-molecule work. The dye can be activated at the carboxy group for coupling purposes.

The **NHS-ester** of the dye reacts easily with amino-groups of proteins and other bio-molecules. Since the amino-group must be non-protonated to be reactive, the pH of the reaction solution has to be adjusted sufficiently high. As with all NHS-esters unavoidable hydrolysis takes place at high pH and competes with the desired labeling. Therefore the solution has to be buffered carefully. For details see the Labeling Protocol on www.atto-tec.com.

The **maleimide** is suitable for labeling sulfhydryl (thiol) groups of proteins, in particular cystein residues. See Labeling Protocol on www.atto-tec.com.

Phalloidin, a bicyclic heptapeptide, is a very strong binding reagent to actin. Fluorescent labeled phalloidin has become a useful tool to investigate the distribution of F-actin within the cytoskeleton of cells by fluorescence microscopy. To prepare a stock solution of the phalloidin-conjugate it is recommended dissolving the sample in 1 ml of methanol.

The **azide** modification is used in the Huisgen reaction ("Click Chemistry").

Further Notes:

- ATTO-TEC products are high-quality reagents intended for research purposes only.
- The use of ATTO-TEC products must be supervised by technically qualified personnel experienced in handling potentially hazardous chemicals. For safety instructions please read the corresponding Material Safety Data Sheet.
- Most ATTO-TEC products and product applications are covered by European and foreign patents.
- Commercial use of ATTO-TEC products is not permitted without written agreement by ATTO-TEC GmbH. Inquiries for licensing may be directed to info@atto-tec.com.